

In the Claims

The following listing of the claims replaces all previous listings.

In the Claims

1. (Currently Amended) A method of preparing an RNA sample substantially free of genomic DNA, comprising the following steps:
 - (a) forming a tissue-lysate/cell lysate from a biological sample;
 - (b) removing essentially all of the gDNA/genomic DNA from the lysate of (a) therein forming a sample preparation;
 - (c) forming an RNA-containing precipitate by adding an organic solvent to the sample preparation of (b);
 - (d) contacting an RNA isolation membrane column with said RNA-containing precipitate of (c), wherein said column comprises a polymeric membrane acting as a physical barrier to said RNA-containing precipitate and retaining the RNA-containing precipitate for purification; and
 - (e) collecting said RNA-containing precipitate from said membrane, wherein said RNA-containing precipitate is substantially free of said genomic DNA.
2. (Canceled)
3. (Currently Amended) The method of claim [[2]] 1, wherein said polymeric membrane is selected from the group consisting of BTSpolysulfone treated with hydroxypropylcellulose, PVDFpoly(vinylidene fluoride), nylon, nitrocellulose, polysulfone, MMMPolysulfone and poly(vinylpyrrolidone), PVPpoly(vinylpyrrolidone), and composites thereof.
4. (Original) The method of claim 1, wherein said membrane has a particle retention ranging from about 0.1 μm to about 10 μm.
5. (Currently Amended) The method of claim 1, wherein said step (b), removing essentially all of the gDNA, is accomplished by using a pre-filtration technique.

6. (Original) The method of claim 1, wherein said lysate is formed employing a lysis buffer comprising a chaotropic agent.
7. (Original) The method of claim 6, wherein said chaotropic agent is selected from a group consisting of guanidine isothiocyanate, ammonium isothiocyanate, guanidine hydrochloride, and combinations thereof.
8. (Original) The method of claim 7, wherein said chaotropic agent is at a concentration ranging from about 0.5 M to about 5.0 M.
9. (Currently Amended) The method of claim 1, wherein said biological sample is selected from the group consisting of animal tissues and plant tissues, animal cells, and/or plant cells.
10. (Currently Amended) The method of claim 9, wherein said biological sample is animal tissues and/or cells are selected from a group consisting of blood, urine, hair, skin, muscle, bone, bodily fluids, and organ extracts and alike.
11. (Currently Amended) The method of claim 1, wherein step (e) is followed by the use of treating the precipitate with DNase treatment.
12. (Original) The method of claim 1, wherein said precipitate comprises RNA essentially free of DNA.
13. (Original) The method of claim 1, wherein said lysate is formed using a lysis buffer comprising β -mercaptoethanol.

14. (Original) The method of claim 1, wherein said organic solvent is an alcohol selected from the group consisting of methanol, ethanol, isopropanol and combinations thereof.

15. (Original) The method of claim 1, wherein said precipitate is washed following step (d) with a wash solution comprising an organic solvent.

16. (Currently Amended) The method of claim 15, wherein said wash solution is selected from the group consisting of ~~Wash Buffer #1 and Wash Buffer #2~~ comprises ethanol and a buffering agent to maintain a pH from about 6 to about 9.

17. (Currently Amended) The method of claim 16, wherein said wash solution Wash Buffer #1, comprises: (a) from about 0.2 to about 2 M guanidine; (b) from about 5 to about 25% ethanol; and (c) a buffering agent to maintain a pH from about 6 to about 9.

18. (Currently Amended) The method of claim 16, wherein said wash solution Wash Buffer #2, comprises: (a) from about 40 to about 90% ethanol; and (b) a buffering agent to maintain a pH from about 6 to about 9.

19. (Currently Amended) A method of preparing an RNA sample substantially free of genomic DNA, comprising the following steps:

- (a) forming a tissue-lysate / cell-lysate from a biological sample;
- (b) contacting a pre-filtration column with said lysate, wherein said pre-filtration column comprises a fiber material, wherein said fiber material has at least one layer of glass or borosilicate fiber; and whereby essentially all genomic DNA in said lysate is removed to produce a filtrate;
- (c) forming an RNA-containing precipitate by adding an organic solvent to said filtrate from step (b);

(d) contacting an RNA isolation membrane column with said RNA-containing precipitate from step (c), wherein said RNA isolation membrane column comprises a polymeric RNA isolation membrane acting as a physical barrier to said RNA-containing precipitate and retaining the RNA-containing precipitate for purification; and

(e) collecting said RNA-containing precipitate from said RNA isolation membrane column, wherein said RNA-containing precipitate is substantially free of said genomic DNA.

20. (Original) The method of claim 19, wherein said fiber material has a particle retention ranging from about 0.1 μm to about 10 μm .

21. (Original) The method of claim 19, wherein said fiber material has a thickness ranging from about 50 μm to about 2000 μm .

22. (Currently Amended) The method of claim [[19]] 21, wherein said fiber material has a specific weight ranging from about 75 g/m^2 to about 300 g/m^2 .

23. (Original) The method of claim 19, wherein said RNA isolation membrane has a particle retention ranging from about 0.1 to about 10 μm .

24. (Currently Amended) The method of claim 19, wherein said RNA isolation membrane is selected from the group consisting of BTSpolysulfone treated with hydroxypropylcellulose, PVDFpoly(vinylidene fluoride), nylon, nitrocellulose, polysulfone, MMMPolysulfone and poly(vinylpyrrolidone), PVPpoly(vinylpyrrolidone), and composites thereof.

25. (Withdrawn) A kit for isolating RNA in a form essentially free from gDNAgenomic DNA, comprising the following: (a) at least one pre-filtration column, wherein said pre-filtration column comprises a fiber material, wherein said fiber material has at least one layer of glass or

borosilicate fiber; (b) at least one RNA isolation membrane column, wherein said membrane column comprises a polymeric membrane; (c) reagents for both (a) and (b); and (d) instructions for isolating RNA with implementing (a) through (c).

26. (Currently Amended) The kit of claim 25, wherein said RNA isolation membrane is selected from the group consisting of BTSpolysulfone treated with hydroxypropylcellulose, PVDFpoly(vinylidene fluoride), nylon, nitrocellulose, polysulfone, MMMPolysulfone and poly(vinylpyrrolidone), PVPpoly(vinylpyrrolidone), and composites thereof.

27. (Withdrawn) The kit of claim 25, wherein said reagents include at least one organic solvent and a lysis buffer.